Supplementary Data

Inactivation of polyunsaturated fatty acid synthesis in the Δ6-fatty acid desaturase deficient (fads2-/-) mouse suppresses atherosclerosis provoked by prolonged high fat / high cholesterol diet

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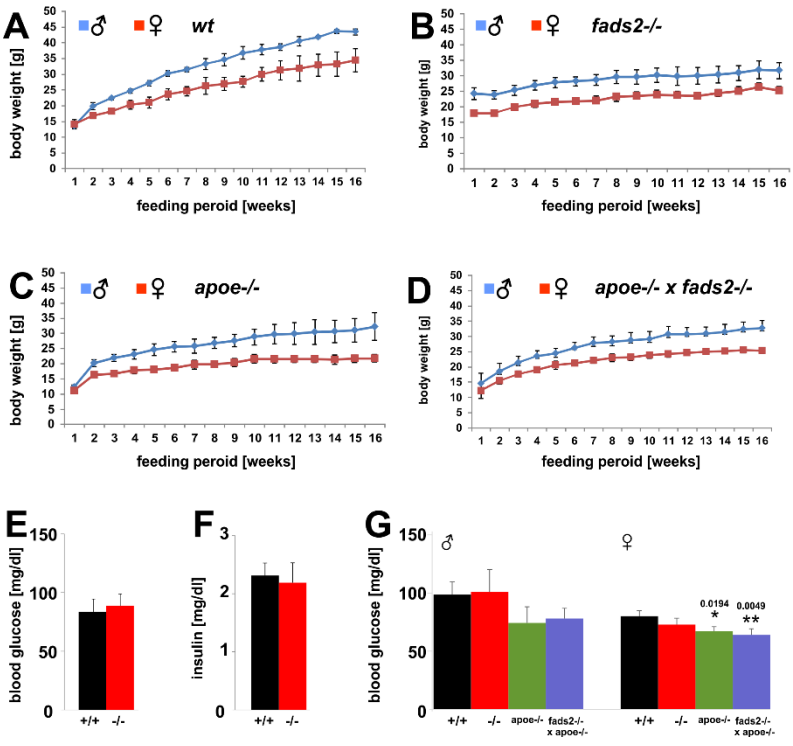
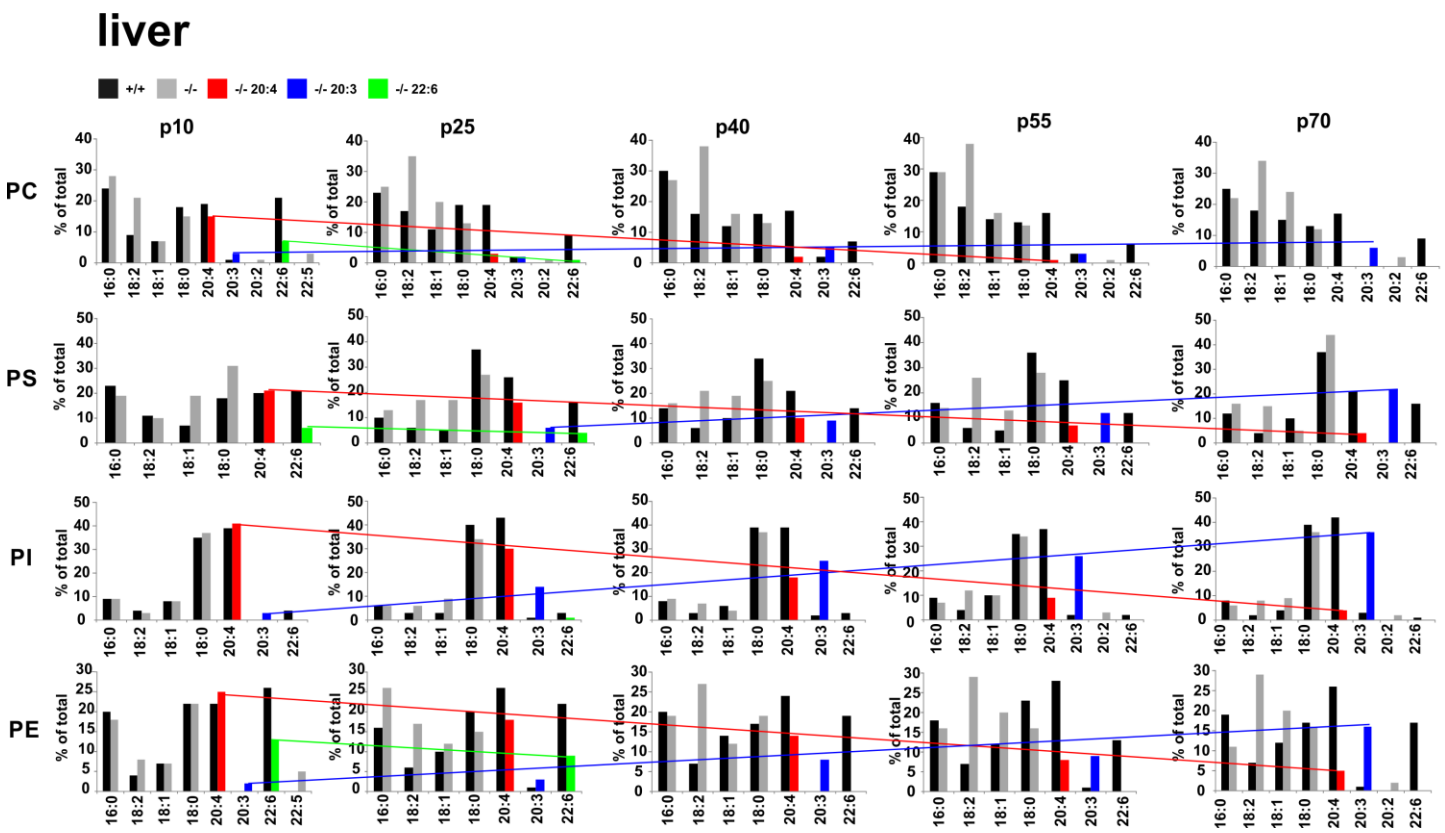
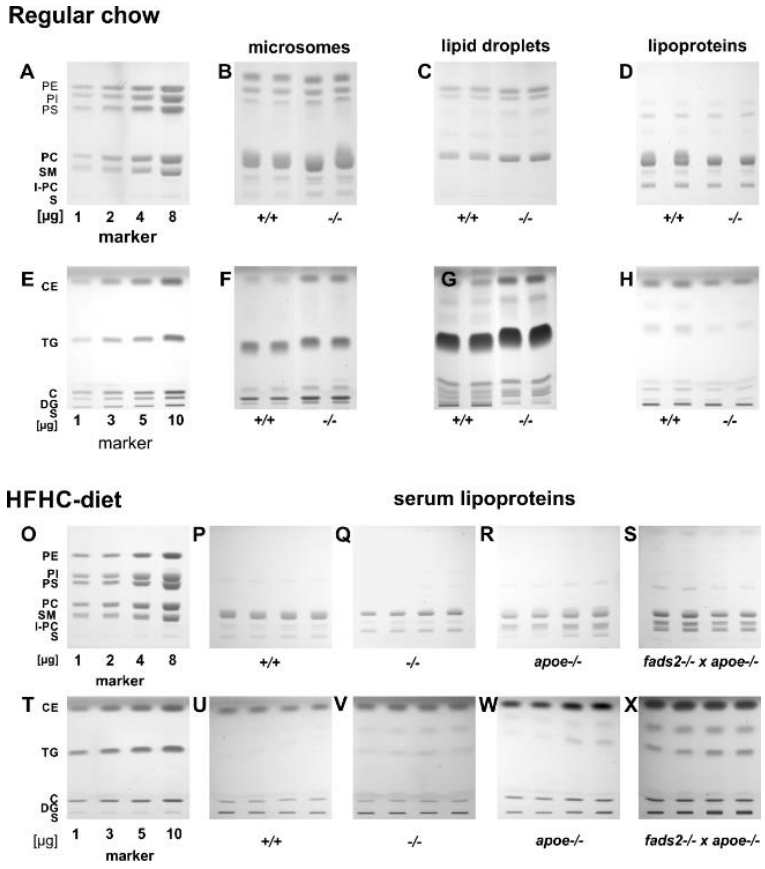


SI Figure 1 Gain of body weight of male and female +/+, *fads2-/-, apoe-/-* and *fads2-/- x apoe-/-* mice during 120-days feeding period of HFHC-diet. Male and female cohorts of (A) +/+, (B) *fads2-/-*, (C) *apoe-/-* and (D) *fads2-/- x apoe-/-* mice.
(E) Blood glucose and (F) insulin concentration of +/+ and *fads2-/- mice on regular chow,* G) Blood glucose concentration of +/+, *fads2-/-, apoe-/-* and *fads2-/- x apoe-/-* male and female mice of over-night starved mice on HFHC-diet.

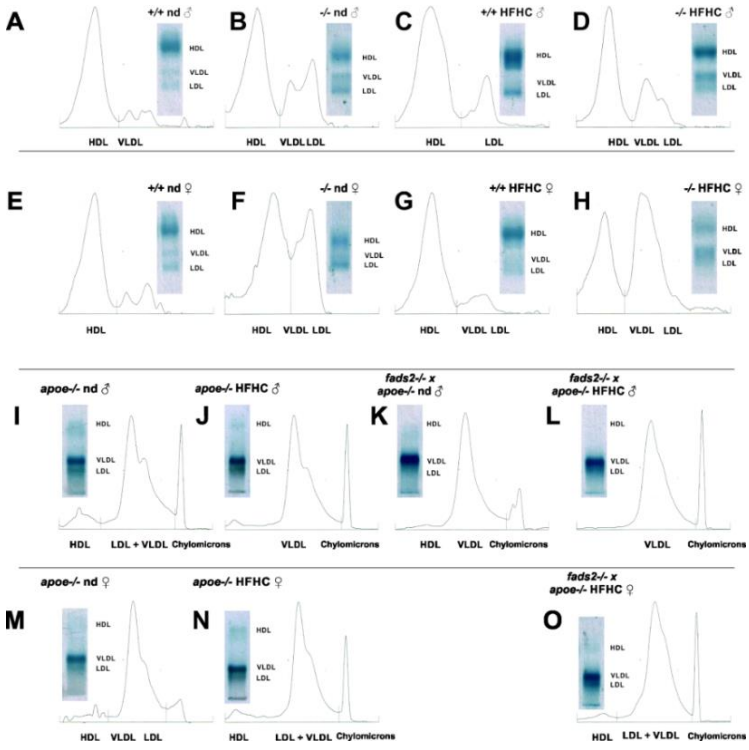


SI Figure 2 Kinetics of the modification of the fatty acid pattern in the phospholipidome of liver of +/+ and fads2-/- mice. HPTLC-separation of PL –classes and densitometric quantification of steady state concentrations of MS/MS-characterized DAG-species of liver phospholipid classes of +/+, fads2-/-, apoe-/- and fads2-/- x apoe-/- mice at p10, p25, p40, p55 and p70.

Fads2-/- liver is depleted from ω3-DHA (green bars) in all PL-classes at p30, ω6-AA (red bars) persisted in trace concentrations and 20:3^{5,11,14} (blue bars) linearly increased to the concentration of AA of the respective PL class, particularly in PS, PI and PS in the developing *fads2-/-* mice.

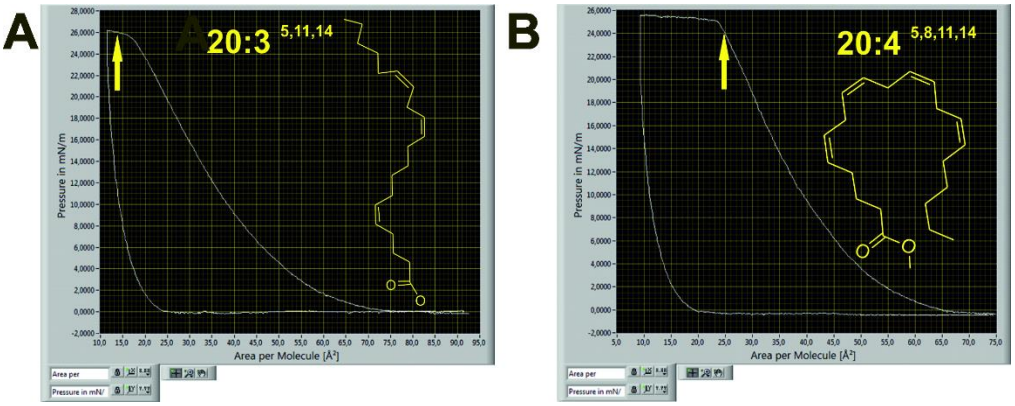


SI Figure 3 Images of densitometric quantification of charred HPTLC-separated PL (B-D) and neutral lipid classes (F-H) in total lipid extracts of liver microsomal fraction, liver lipid droplets and lipoproteins. Quantification PL (P-S and NL (U-X) of serum lipoproteins) after 120 days HFHC-diet of +/+ and *fads2-/- apoe-/-* and *fads2-/- x apoe-/-* mice after 120 days normal chow. Lipid extracts of four mice each were pooled for three analyses each. (A, D, O, T) marker lipids.



SI Figure 4 HFHC-diet causes severe changes of Lp – profiles of male and female *fads2-/-* mice. Sebia HYDRASYS agarose gel electrophoresis Quantification the serum lipoprotein pattern of adult (4mo) mice on regular chow of (A) male +/+ and (E) female +/+, (B) and (F) *fads2-/-,* and (I) and (M) *apoe-*/-,(K) *fads2-/- x apoe-/-* male; on HFHC-diet (C) and (G) +/+ and (D) and (H) *fads2-/-,* (J) and (N) *apoe-/-* and (L) and (O) *fads2-/- x apoe-/-*. Cohort sizes n=3.

π-A- isotherms were recorded continuously and automatically. The teflon coated thermostated trough with the sub-phase was kept at constant temperature of 25°C. Benzene was used as solvent for spreading the monolayer on double distilled water. The synthesis and properties of 20:3^{5,11,14}, synthesized in this laboratory, have been described previously [1].
Pressure at the collapse point of 20:3^{5,11,14} was 26.00mN/m and the molecular area 12Å^2 compared to 25.00mN/m and 22.5 Å^2 of 20:4 ^{5,8,11,14}.



SI Figure 5 25°C π-A- isotherms of 20:3^{5,11,14} and 20:4^{5,8,11,14} .The properties of the monolayers of 20:3^{5,11,14} and 20:4^{5,8,11,14} were studied with the RK1-Standard horizontal Langmuir-type surface film balance with full analog electronic to measure pressure, area, barrier speed, temperature and data recording by computer (Riegler & Kirstein,14467 Potsdam, Germany).

Reference

[1] Hammels, I., Binczek, E., Schmidt-Soltau, I., Jenke, B., Thomas, A., Vogel, M., et al., 2019. Novel CB1-ligands maintain homeostasis of the endocannabinoid-system in omega3- and omega6-long chain-PUFA deficiency. J Lipid Res. 60(8):1396-1409